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SUB-HAPLOID POLLEN IN *AMARANTHUS*

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AS pointed out by Khoshoo<sup>1</sup> a study of pollen mitosis offers several advantages, one of the important being the study of chromosome balance and fertility in numerical hybrids. Some interesting features came to light during the course of such an investigation on a dibasic triploid *Amaranthus* hybrid ( $2n=49$ ) reported earlier by Pal and Khoshoo.<sup>2</sup>

The hybrid arises spontaneously wherever *A. dubius* ( $2n=64$ ,  $x=16$ ) and *A. spinosus* ( $2n=34$ ,  $x=17$ ) grow in sufficient proximity. The parents have normal meiosis with 32 and 17 chromosomes (Figs. 1-2) respectively in pollen grains. Pollen of the amphiploid, *A. dubio-spinosus* ( $2n=98$ ), contains 49 chromosomes (Fig. 3). In all these the pollen is shed at trinucleate stage.

More than 50% pollen mother cells of the  $F_1$  *A. dubius*  $\times$  *spinosus* ( $2n=49$ ) contain 17 II + 15 I. The two extremes, however, are 19 II + 11 I and 16 II + 17 I. Pollen of the hybrid is of two types, macro- (21-26  $\mu$ , 77%) and micropollen (8-14  $\mu$ , 23%). Both occur regularly. Lagging univalents are the source of micropollen. Chromosome number in the former ranges from 19 to 27 (Figs. 4-5), while the latter contains only 1 to 4 chromosomes (Figs. 6-8). Among micropollen grains, those containing one chromosome are the most common (44%), followed by those with two (39%), four (11%) and three (6%) chromosomes. Although all such grains are much smaller than macropollen, yet the two appear to be alike in external character of exine. There is a general correlation between number of chromosomes and size of pollen, but increase or decrease by about four chromosomes is unable to affect the size.

A distinct difference between the rate of development in the two types of pollen (Fig. 10) has been noted. However, there is no difference at prophase, but the same is apparent at metaphase. Furthermore, micropollen does not proceed beyond this stage, but all macropollen becomes binucleate following an asynchronous development. Only 8% macropollen is trinucleate at anthesis. The balanced grains probably develop faster than the unbalanced ones. This asynchrony, found between and within micro- and macropollen, naturally has been created by qualitative and quantitative segregation at meiosis. Possibly there may also be associated differences in cytoplasm.

The total absence of anaphase and binucleate condition in micropollen is perhaps due to the absence of spindle formation. However, chromosomes become double (Figs. 6-7) indicating a normal cycle of DNA synthesis. Since the spindle is composed largely of protein and some RNA, it appears that quantity, particularly of the former, sufficient to organize a normal spindle is not synthesized during the resting stage. This point needs experimental verification. The net result is that directive influence of spindle on centromeres of otherwise double chromosomes is absent.

The strong developmental difference between the two types of pollen (Fig. 10) vanishes altogether when due to cold shock, walls either fail to develop or are feebly developed among the constituents of a tetrad ensuing from a single pollen mother cell (Fig. 9). In such cases there is synchrony not only between macro- and micropollen within a single tetrad, but also at intertetrad level. The former is perfectly understandable, because components of one pollen mother cell form a single balanced unit as long as they are held together, thereby enabling a free exchange of metabolites. The loss in one component compensates for the gain in another. Under such circumstances deficient nuclei can develop at the same rate as others. The intertetrad synchrony or developmental co-ordination within an anther sac appears also to be the result of feeble wall formation; maybe cytotoxic channels<sup>8</sup> persist.

It is clear that in *Amaranthus* ( $x=16$  and 17) pollen grains containing a single chromosome are deficient to the extent of 94%, and they are produced regularly in the  $F_1$  hybrid. This is a unique situation because it is well known that pollen lacking just one chromosome in a basic complement ( $x-1$ ), does not generally develop independently.<sup>4-7</sup> However, two interesting cases have been reported previously. Johnsson<sup>9</sup> made a passing reference about the occurrence of grains with one to seven chromosomes in a polymitotic form of *Alopecurus* ( $x=7$ ), but nothing more is known about it. Only two (out of 298) pollen grains with  $n=3$ , deficient to the extent of 50%, were observed in *Tradescantia* ( $x=6$ ).<sup>4</sup> Obviously in *Amaranthus* a single chromosome is reasonably balanced and, what is most important, survives independently for some time, enables external development of apparently normal exine,

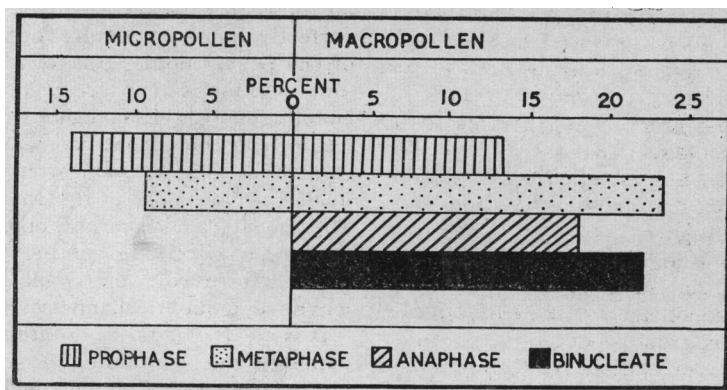
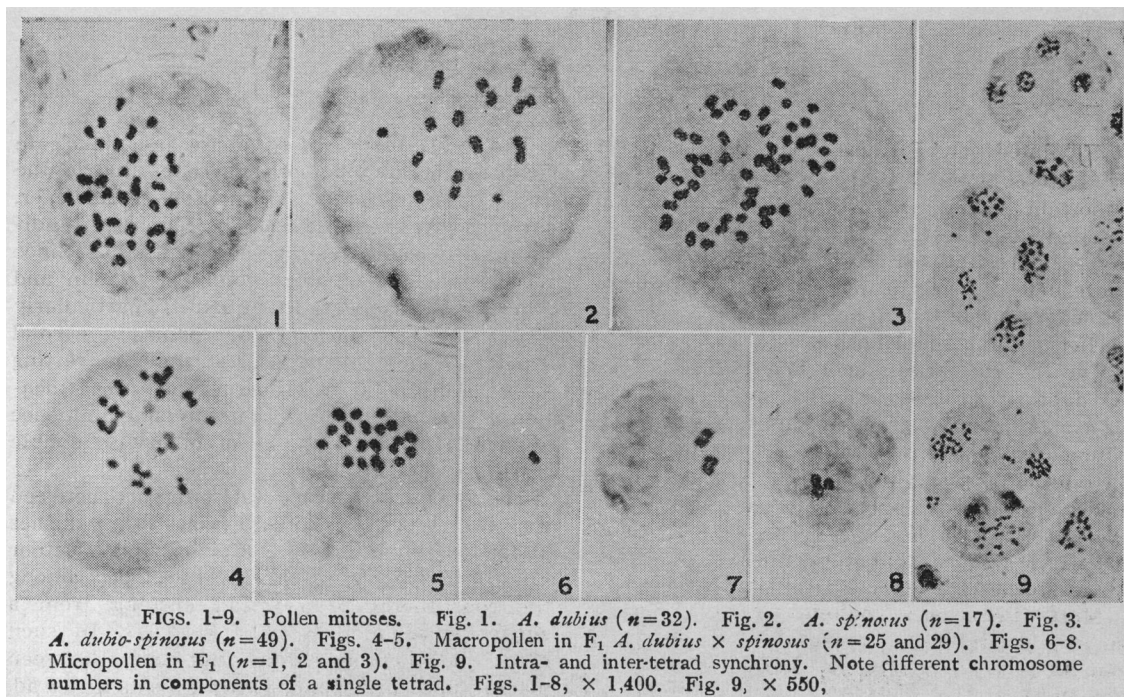


FIG 10. Histogram of the relative frequency and development of micro- and macropollen in the  $F_1$  hybrid.

internally completes DNA synthesis, and finally proceeds up to metaphase. However, a cytogenetic study of the progeny of the  $F_1$  hybrid reveals that micropollen is ineffective in fertilization.

*Amaranthus* offers a fine material in which cells with the lowest possible chromosome number ( $n=1$ ) can exist up to a certain stage. If the life of such cells ( $n=1, 2, 3, 4$ ) could be prolonged *in vitro* and induced to form a tissue, one would have an excellent experimental material for an intensive study of some of the fascinating problems in cell biology.

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